



## **USDA ARS National Animal Germplasm Program**

### **Bull Semen Collection and Transportation**

#### **Supplies that will be sent to the cooperator:**

- Impact Shipper with instructions for use (see Impact Shipper Protocol on the Animal GRIN webpage: <https://www.ars.usda.gov/plains-area/fort-collins-co/center-for-agricultural-resources-research/paagrpru/docs/animal/animal-protocols/>);
- Cooling packs;
- Sample tubes, 15 or 50 mL;
- Zip lock bags;
- Frozen Tris-Egg yolk A (TCA) Cooling media
- Antibiotics, sterile water and syringes

#### **Prior to collection**

Thaw the frozen media and warm it to 37 °C. Maintain it at this temperature for all dilutions. Ensure that the cooling packs are at 5 °C (**not frozen but at 5 °C**).

#### **Collection Process**

1. Label a sample tube with the bull's name and/or identification number.
2. Collect semen from sexually mature bulls via an artificial vagina or electroejaculation.
3. Check sample to ensure it is free of urine and other contaminants and maintain the sample at 35 to 37 °C.
4. Determine the sample volume, sperm concentration, and total motility.
5. Multiplying the two (volume x concentration) will provide the sperm count.
6. Divide the sperm count by  $120 \times 10^6$  sperm/mL which will provide the final dilution volume (sample plus TCA).
7. The final volume minus the sample volume will determine the amount of the 37 °C TCA cryopreservation media to add to the sample.
8. Dilute the sample with 37 °C TCA cryopreservation media as determined in step 6.
9. Place the sealed tube in a refrigerator or 5 °C environment.
10. After collection, dilution and cooling of all semen samples, pack the Impact Shipper with the samples according to the instructions in the Impact Shipper Protocol (<https://www.ars.usda.gov/plains-area/fort-collins-co/center-for-agricultural-resources-research/paagrpru/docs/animal/animal-protocols/>) and seal the box.

Dilution math example:

Sample volume (VOL):	4.5 mL
Sperm concentration (CONC):	$300 \times 10^6$ sperm/mL
Sperm count (VOL X CONC):	$1350 \times 10^6$ sperm/mL
Final diluted volume (count $\div$ 120 x $10^6$ sperm/mL):	11.25 mL
Amount of TCA to add to the sample (Final volume less sample volume):	6.75 mL

Versions: October 2019, April 2020